CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

Saflufenacil

Chemical Code #5977, **Tolerance** # 53057
SB 950 # NA

9/30/08

I. DATA GAP STATUS

Chronic toxicity, rat:	No data gap, no adverse effect indicated
Chronic toxicity, dog:	No data gap, no adverse effect indicated
Oncogenicity, rat:	No data gap, no adverse effect indicated
Oncogenicity, mouse:	No data gap, no adverse effect indicated
Reproduction, rat:	No data gap, no adverse effect indicated
Teratology, rat:	No data gap, no adverse effect indicated
Teratology, rabbit:	No data gap, no adverse effect indicated
Gene mutation:	No data gap, no adverse effect indicated
Chromosome effects:	No data gap, possible adverse effect indicated
DNA damage:	No data gap, no adverse effect indicated
Neurotoxicity:	Study not required at this time.

Toxicology one-liners are attached.

All record numbers through 237981 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.
indicates a study on file but not yet reviewed.
File name: T080930
Revised by T. Moore, 9/30/08

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may contain additional effects.

COMBINED, RAT

** 53057-0033; 237966; "BAS 800 H - Combined Chronic Toxicity/Carcinogenicity Study in Wistar Rats; Administration Via the Diet up to 24 Months"; (U. Kaspers, V. Strauss, S. Groters, R. Fabian, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, FRG; Report No. 80S0414/01170; 12/13/07); Fifty Wistar rats/sex/group (unless otherwise noted) received 0, 20, 100, 250 (males only), 500 or 1000 (females only) ppm of BAS 800 H (batch no. COD-000515, purity: 93.8%) in the diet for 24 months ((M) 0, 0.9, 4.8, 12.0, 24.2, (F) 0, 1.3, 6.2, 31.4, 63.0 mg/kg/day). Survival of the study animals was not affected by the treatment. The mean body weights of the males in the 500 ppm group were less than those of the control group during the first 3 months of the study ($p<0.05$). Thereafter, a statistically-significant effect was not observed. No treatment-related effect upon the mean food consumption was evident. The ophthalmology examination did not reveal any treatment-related effects. In the hematology evaluation, the mean hemoglobin concentrations, mean hematocrit, mean corpuscular volume and mean corpuscular hemoglobin values of the males in the 500 ppm group and the females in the 1000 ppm group were less than the control values at some time point during the 1st year of treatment ($p<0.01$ or 0.05). The mean hematocrit of the 250 ppm males and the 500 ppm females was also less than the control values for at least one time point during the 1st year ($p<0.01$ or 0.05). The hemoglobin concentration of the 500 ppm females was less than the control value after 6 months of treatment ($p<0.01$). In the clinical chemistry evaluation, the total protein and albumin concentrations in the serum of the 500 ppm males were less than the control values after 6 months of treatment ($p<0.01$ or 0.05). The mean total protein value for the 250 ppm males were also less than that of the control group at the same time point ($p<0.05$). The females in the 1000 ppm group demonstrated increased levels of alanine aminotransferase and alkaline phosphatase activities in the serum at some time point(s) during the 1st year ($p<0.01$ or 0.05). The 500 ppm females also had elevated alanine aminotransferase levels in the serum throughout the 1st year (NS, $p<0.05$). In the urinalysis, the urobilinogen levels were elevated for the 250 and 500 ppm males at 3 and 12 months ($p<0.01$) and for the 500 and 1000 ppm females at 3 months ($p<0.01$ or 0.05). In the necropsy examination, the mean absolute liver and spleen weights of the males in the 500 ppm group (main study) were less than the control values ($0<0.01$). However, the mean relative liver and spleen weights of these animals were not affected and no histological lesions were evident in these organs. The mean relative epididymidal weights of the 250 and 500 ppm males (main study) were greater than the control value ($p<0.01$ or 0.05). However, no lesions were evident in this organ as well. In the histological examination, no treatment-related lesions were noted. **No adverse effect indicated. Rat Chronic Dietary Toxicity NOEL:** (M/F) 100 ppm ((M) 4.8 mg/kg/day, (F) 6.2 mg/kg/day) (based upon the treatment-related effects on hematology parameters and the presence of higher urobilinogen content in the urine of the 250 ppm males and the 500 ppm females); **no oncogenicity evident. Study acceptable.** (Moore, 6/13/08)

CHRONIC TOXICITY, RAT

See Combined Rat above.

CHRONIC TOXICITY, DOG

** 53057-0031; 237964; "BAS 800 H - Chronic Oral Toxicity Study in Beagle Dogs, Administration via Gelatin Capsules for 12 Months"; (K. Hempel, V. Strauss, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 43D0414/01196; 11/14/07); Five beagle dogs/sex/group received 0, 5, 20, or 80 mg/kg/day of BAS 800 H (batch no. COD-000515, purity: 93.8%) orally, in capsules, for 52 weeks. The mean body weight gain and food consumption data did not demonstrate a treatment-related effect. Four males and three females in the 80 mg/kg group had dark brown coloration of their feces between study days 9 and 27. The ophthalmological examination and urinalysis did not reveal any treatment-related effects. In the hematological evaluation, the mean corpuscular volume and corpuscular hemoglobin values of both sexes in the 80 mg/kg group were less than

those of the controls throughout the study (p<0.01). In the clinical chemistry evaluation, the serum alkaline phosphatase activities of both sexes in the 80 mg/kg group were elevated above the control values throughout the study (NS, p<0.01). The serum albumin concentrations of both sexes in the 80 mg/kg group were less than those of the controls throughout the study (NS, p<0.01). Increased iron storage (grade $\geq$ 2) was noted in the livers of both sexes in the 80 mg/kg group and the males in the 20 mg/kg group ((M) 0: 0/5 vs. 20: 3/5, 80: 3/5; (F) 0: 1/5 vs. 80: 4/5); **No adverse effect indicated. Dog Chronic Oral Toxicity NOEL: (M)** 5 mg/kg/day (based upon the increased level of iron storage in the liver of the 20 mg/kg males), **(F)**: 20 mg/kg/day (based upon treatment-related effects noted in the hematology evaluation and increased iron storage in the liver of the 80 mg/kg females). **Study acceptable**. (Moore, 5/28/08)

ONCOGENICITY, RAT

See Combined Rat above.

ONCOGENICITY, MOUSE

** 53057-0032; 237965; "BAS 800 H - Carcinogenicity Study in C57BL/6NCrL Mice, Administration via the Diet over 18 Months"; (H. Kamp, V. Strauss, K. Kuttler, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 87C0414/01177; 12/5/07); Fifty C57BL/6NCrL mice/sex/group (unless otherwise noted) received 0, 1 (males only), 5, 25, 75 or 150 (females only) ppm of BAS 800 H (batch no. COD-000515, purity: 93.8%) in the diet for up to 18 months (note: due to the high mortality for the males in the control and 5 ppm group, the surviving males in the study were euthanized on study days 527 and 528, 20 days prior to the scheduled sacrifice) ((M) 0, 0.2, 0.9, 4.6, and 13.8 mg/kg/day, (F) 0, 1.2, 6.4, 18.9, 38.1 mg/kg/day). An additional 10 animals/sex/group were included in the control, the 75 ppm male and the 150 ppm female groups and were treated for 10 months prior to being euthanized. The mean body weights of the males in the 75 ppm group and the females in the 25, 75 and 150 ppm groups were less than the control values during the first month of the study (p<0.01). By the 2nd month of the study, no treatment-related effect was evident. There was no apparent treatment-related effect on the mean food consumption. In the hematology evaluation for the animals in the satellite group after 10 months of treatment, the mean red blood cell count, hemoglobin concentration and hematocrit of the 75 ppm males and the 150 ppm females were less than the control values (p<0.01 or 0.05). No treatment-related effect was noted in the differential white blood cell count at any of the time points evaluated. The total poryphyrin content in the feces and liver of the 75 ppm males and the 150 ppm females in the satellite group was greater than the control values (p<0.01). In the necropsy examination, the mean relative liver weight of the males in the 75 ppm satellite group was greater than that of the control (p<0.05). However, no treatment-related lesions were noted in the livers of these animals. In the histopathology examination of the animals in the main study, lipogenic pigment was noted in the livers of the 25 and 75 ppm males (0: 0/50 vs. 25: 9/50, 75: 37/50). An increased incidence of karyomegaly was also noted in the livers of these animals (0: 2/50 vs. 25: 6/50, 75: 16/50). No treatment-related lesions were evident for the females in the histological examination. **No adverse effect indicated. Mouse Chronic Toxicity NOEL: (M)** 5 ppm (0.9 mg/kg/day) (based upon the lesions noted in the livers of the 25 ppm males), **(F)** 75 ppm (18.9 mg/kg/day) (based upon the treatment-related effects on the hematology and the greater total porphyrin concentration in the feces and livers of the 150 ppm females); **no oncogenicity was evident. Study acceptable.** (Moore, 6/10/08)

REPRODUCTION, RAT

** 53057-0036; 237969; "BAS 800 H - Two-Generation Reproduction Toxicity Study in Wistar Rats, Administration Via the Diet"; (S. Schneider, V. Strauss, K. Kuettler, E. Fabian, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 70R0414/01200; 12/19/07); Twenty five Wistar rats/sex/group were treated in the diet with 0, 5, 15, or 50 mg/kg/day of BAS 800 H (batch no. COD-000515, purity: 93.8%) for 2 generations. The treatment period for the F0 generation included at least 10 weeks prior to mating, the mating period, and 3 weeks each for gestation and lactation (F1 litter). Twenty five rats/sex/group were selected from the F1 litter and treated for at least 10 weeks prior to mating, during the mating period and an additional 3 weeks each for

gestation and lactation (F2 litter). The F0 and F1 parental females in the 50 mg/kg group demonstrated lower mean body weight gain than did the control animals during the gestation period ($p<0.01$). The F1 parental males in the 50 mg/kg group exhibited lower mean body weight gain during the premating period ($p<0.01$). The mean food consumption of the 50 mg/kg females in both generations was less than that of the control females during the lactation period ($p<0.01$). In the hematology evaluation, the mean hemoglobin concentration, the hematocrit, mean corpuscular volumes and mean corpuscular hemoglobin of both sexes in the 50 mg/kg group of both generations were less than the control values ($p<0.01$). The hemoglobin concentration, hematocrit and mean corpuscular volume of the 15 mg/kg males in both generations were less than the control values ($p<0.01$ or 0.05). The mean corpuscular hemoglobin concentrations of the 50 mg/kg males in both generations were also less than the control values ($p<0.01$). Although other parameters were statistically different from the control values, none of them demonstrated a consistent effect through out the two generations. In the clinical chemistry evaluation, the mean total serum protein concentrations of both sexes in the 50 mg/kg group in the F0 generation and of the 50 mg/kg males in the F1 generation were less than those of the control ($p<0.01$). The mean serum albumin and globulin values of the 50 mg/kg males in both generations were less than the control values ($p<0.01$ or 0.05). Although other parameters were statistically different from the control values, none of them demonstrated a consistent effect through out the two generations. In the necropsy of the parental animals, the mean absolute and relative spleen weights of the males in the 50 mg/kg group of both generations were greater than the control values ($p<0.01$ or 0.05). Although the mean relative weights of the testes, epididymides, and cauda epididymis of the 50 mg/kg males in the F1 generation were greater than the control values, no histopathological lesions were evident in these tissues. Likewise, the mean absolute and relative uterine weights of the 50 mg/kg females in the F0 generation were less than the control group. However, no lesions were noted in the histopathological examintion. There was no treatment-related effect upon the fertility or gestation indices. The mean litter sizes of the 50 mg/kg group in both generations was less than the size of the control litters. The viability index of the pups in the 50 mg/kg group of both generations was less than the control index. Developmentally, the 50 mg/kg pups of both generations had lower mean body weights from day 1 post-partum ($p<0.05$ or $p<0.01$). **No adverse effect indicated. Parental NOEL:** 5 mg/kg/day (based upon the treatment-related effects upon the hematology parameters of the males in the 15 mg/kg group); **Reproductive NOEL:** 15 mg/kg/day (based upon the smaller litter sizes of the dams in the 50 mg/kg group of both generations); **Developmental NOEL:** 15 mg/kg/day (based upon the lower mean body weights of the 50 mg/kg pups in both generations). **Study acceptable.** (Moore, 7/8/08)

TERATOLOGY, RAT

** 53057-0034; 237967; "BAS 800 H - Prenatal Developmental Toxicity Study in Wistar Rats, Oral Administration (Gavage)"; (S. Schneider, V. Strauss, W. Kaufmann, E. Fabian, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, FRG; Report No. 30R0414/01178; 1/25/07); Twenty five time-mated female Wistar rats/group were dosed orally by gavage with 0 (vehicle: aqueous 1.0% carboxymethylcellulose), 5, 20, or 60 mg/kg/day from gestation day 6 through gestation day 19 with BAS 800 H (batch no. COD-000515; purity: 93.8%). All of the study animals survived the treatment period. There was no treatment-related effect upon the mean body weights or food consumption of the dams. In the hematology evaluation, the mean hematocrit values of the 20 and 60 mg/kg dams were less than that of the control ($p<0.05$). The mean hemoglobin, mean corpuscular volume and mean corpuscular hemoglobin of the 60 mg/kg females were less than the control values ($p<0.01$ or 0.05). The total porphyrin in the livers of the 5 mg/kg females and above was greater than that of the control ($p<0.01$). The mean body weights of the 20 and 60 mg/kg fetuses were less than the control values ($p<0.01$ or 0.05). There was an increased incidence in skeletal malformations in the litters of the 60 mg/kg group (0: 1/22 vs. 60: 7/22, $p<0.05$). Among the 10 fetuses suffering skeletal malformations, the following malformed bones were noted with the incidences in parentheses: bent scapula (5), malpositioned and bipartite sternebra (2), thick humerus (5), bent radius (4), bent ulna (3), and bent femur (3). **No adverse effect indicated. Maternal NOEL:** < 5 mg/kg/day (based upon the increased levels of total porphyrin in the liver of the 5 mg/kg dams);

Developmental NOEL: 5 mg/kg/day (based upon the reduced mean body weights of the 20 mg/kg fetuses). **Study acceptable.** (Moore, 6/17/08)

TERATOLOGY, RABBIT

** 53057-0035; 237968; " BAS 800 H - Prenatal Developmental Toxicity Study in Himalayan Rabbits, Oral Administration (Gavage); (S. Schneider, K. Deckhardt, J. Hellwig, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, FRG; Report No. 40R0414/01173; 2/8/06); Twenty five artificially inseminated female Himalayan rabbits were dosed orally by gavage with 0 (vehicle: aqueous 1.0% carboxymethylcellulose), 50, 200 or 600 mg/kg/day of BAS 800 H (batch no. COD-000515; purity: 93.8%) from gestation day 6 through gestation day 28. In the 600 mg/kg group, four animals suffered abortions, two were euthanized in moribund condition and one died prematurely. One female in the 50 mg/kg group suffered an abortion and one female in the 200 mg/kg group died during the study. The mean food consumption of the does in the 600 mg/kg group was less than the control group during the first days of dosing ($p<0.05$). There was no apparent effect on the mean body weight gain of these does. No treatment-related effects were noted for the hematology parameters. The mean total porphyrin content in the livers of the 50 mg/kg does and above was greater than that of the control group ($p<0.01$). The mean total porphyrin levels in the fetal livers of both sexes in the 200 and 600 mg/kg groups were greater than the control values ($p<0.01$ or 0.05). **No adverse effect was evident. Maternal NOEL:** <50 mg/kg/day (based upon the increased total porphyrin levels in the liver of the 50 mg/kg does); **Developmental NOEL:** 50 mg/kg/day (based upon the increased total porphyrin levels in the livers of the 200 mg/kg fetuses); **Study acceptable.** (Moore, 6/18/08)

GENE MUTATION

** 53057-0037; 237970; "*Salmonella typhimurium/Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 800 H"; (G. Englehardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 40M0414/014210; 6/17/05); *Salmonella typhimurium* strains TA1535, TA1537, TA98 and TA100 and *Escherichia coli* strain WP2 uvrA were tested with concentrations of BAS 800 H (batch no. COD-000515, purity: 93.8%) ranging from 55 to 5500 ug/plate for induction of mutations in 2 trials with and without rat liver activation. In the 1st trial, the strains were exposed to the test material, which had been plate incorporated for 48 to 72 hours at 37° C. In the 2nd trial, the strains were preincubated with the test material for 20 minutes prior to being incubated for 48 to 72 hours, using the plate incorporation method. There were triplicate plates per treatment level. An Aroclor 1254-induced rat liver S9 fraction was used to metabolize the test material. **No adverse effect indicated.** There was no increase in the incidence of revertant colonies under conditions of either activation or non-activation. Positive controls were functional. **Study acceptable.** (Moore, 7/8/09)

** 53057-0037; 237971; "*In Vitro* Gene Mutation Test with BAS 800H in CHO Cells (HPRT Locus Assay)"; (G. Engelhardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 50M0414/014213; 9/28/05);
Chinese Hamster Ovary cells (CHO, substrain K1) were exposed to concentrations of BAS 800 H (batch no. COD-000515; purity: 93.8%) ranging from 312.5 to 5000 ug/ml for 4 hours at 37° C under conditions of activation and non-activation in two trials. Duplicate cultures were performed for each treatment level. The S9 fraction used to metabolize the test material was derived from the livers of male Sprague-Dawley rats pretreated with Aroclor 1254. An increased rate of mutation was not evident at any treatment level with or w/o activation. **No adverse effect indicated.** The positive controls were functional. **Study acceptable.** (Moore, 7/11/08)

** 53057-0042; 237981; "*Salmonella typhimurium/Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 800 H - Anhydrate"; (G. Englehardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 40M0267/054031; 11/17/05); *Salmonella typhimurium* strains TA1535, TA1537, TA98 and TA100 and *Escherichia coli* strain WP2 uvrA were tested with concentrations

of BAS 800 H - anhydrate (batch no. 31896/082, purity: 99.0%) ranging from 20 to 5000 ug/plate for induction of mutations in 2 trials with and without rat liver activation. In the 1st trial, the strains were exposed to the test material, which had been plate incorporated, for 48 to 72 hours at 37° C. In the 2nd trial, the strains were preincubated with the test material for 20 minutes prior to being incubated for 48 to 72 hours, using the plate incorporation method. There were triplicate plates per treatment level. An Aroclor 1254-induced rat liver S9 fraction was used to metabolize the test material. **No adverse effect indicated.** There was no increase in the incidence of revertant colonies under conditions of either activation or non-activation. Positive controls were functional. **Study acceptable.** (Moore, 7/29/09)

CHROMOSOME EFFECTS

**** 53057-0037; 237972**; "*In Vitro* Chromosome Abberation Assay with BAS 800H in V79 Cells"; (G. Engelhardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 32M0414/014214; 10/19/05); V79 Chinese hamster cells were exposed to concentrations of BAS 800 H (batch no. COD-000515; purity: 93.8%) ranging from 156.25 to 5000 ug/ml under conditions of activation and non-activation for 4 hours and incubated for an additional 14 hours in the 1st trial. In the 2nd trial, the non-activated cultures were exposed to the test material at concentrations ranging from 250 to 4000 ug/ml for 18 hours and then were either harvested or exposed to concentrations of the test material ranging from 1000 to 4000 ug/ml for 18 hours and incubated for an additional 10 hours. The activated cultures were exposed to concentrations of the test material ranging from 1000 to 5000 ug/ml for 4 hours and incubated for an additional 24 hours. In the 3rd trial, the activated cultures were exposed to concentrations of the test material ranging from 500 to 4000 ug/ml for 4 hours and incubated for an additional 24 hours. An Arochlor 1254-induced rat liver S9 fraction was used to metabolize the test material. Duplicate cultures were performed at each treatment level. One hundred metaphases per culture were evaluated (200 metaphases per treatment level) (only 100 metaphases per treatment level for the positive control samples). There was a treatment-related increase in chromosomal cell aberrations under conditions of activation. Positive controls were functional. **Adverse effect indicated. Study acceptable.** (Moore, 7/17/08)

DNA DAMAGE

** 53057-0037; 237973; "*In Vivo* Unscheduled DNA Synthesis (USD) Assay with BAS 800H in Rat Hepatocytes, Single Oral Administration'; (G. Engelhardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 80M0414/014212; 9/29/05); Six Wistar male rats/group received a single oral dose of 0 (aqueous 0.5% carboxymethyl cellulose), 1000 or 2000 mg/kg of BAS 800 H (batch no. COD-000515; purity: 93.8%) by gavage. Hepatocytes from 3 animals/group/time point were isolated at 3 and 14 hours post-dose. Viability was determined by trypan blue dye exclusion and ranged from 90.3 to 94.3%. After attachment, cells were exposed to (methyl-^{3}H) thymidine for 4 hours followed by 12-hour incubation with unlabelled thymidine. One hundred cells were scored per animal (2 to 3 slides). No increase in the net nuclear grain counts was evident at any dose level or sampling time. **No adverse effect indicated**. The positive control was functional. **Study Acceptable**. (Moore, 7/17/08).

** 53057-0037; 237974; "Cytogenetic Study *In Vivo* with BAS 800H in the Mouse Micronucleus Test, Single Oral Administration"; (G. Engelhardt, E. Leibold; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 26M0414/014211; 8/11/05); Five male NMRI mice/group were dosed orally by gavage with 0 (aqueous 0.5% carboxymethyl cellulose), 500, 1000 or 2000 mg/kg of BAS 800 H (batch no. COD-000515; purity: 93.8%). The animals were euthanized 24 hours post-dose. An additional cohort of 5 males/group were dosed with 0 or 2000 mg/kg of the test material and euthanized at 48 hours post-dose. Positive control groups of five males/group were dosed with either 20 mg/kg of cyclophosphamide orally by gavage or 0.15 mg/kg of vincristine intraperitoneally and were euthanized at 24 hours post-dose. The femoral bone marrow was harvested and evaluated for the presence of micronuclei in both polychromatic and normochromatic erythrocytes. Two thousand polychromatic erythrocytes were evaluated per animal. There was no treatment-related

increase in the number of micronuclei per 2000 polychromatic erythrocytes. The positive controls were functional. **No adverse effect indicated. Study acceptable.** (Moore, 7/18/08)

NEUROTOXICITY

Rat Acute Neurotoxicity

53057-0022; 237921; "BAS 800 H - Acute Oral Neurotoxicity in Wistar Rats; Administration via Gavage (Including Amendment No. 1)"; (U. Kaspers, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 61S0414/01208; 8/27/07); Ten Wistar rats/sex/group were dosed orally by gavage with 0, 125, 500 and 2000 mg/kg of BAS 800 H (batch no. COD-000515; purity: 93.8%). The vehicle was aqueous 0.5% carboxymethyl cellulose. No deaths resulted from the treatment. No treatment-related clinical signs were revealed in the functional observational battery. The mean overall activity of the 2000 mg/kg males was reduced on day 0 ($p<0.05$). No treatment-related lesions were observed in the neuropathology evaluation. **No adverse effect indicated. Reported Acute Neurotoxicity NOEL:** (M) 500 mg/kg (based on reduced motor activity of the 2000 mg/kg males) (F) > 2000 mg/kg; **Study unacceptable**, possibly upgradeable to acceptable with concurrent positive control study data**.** (Moore, 5/8/08)

Rat Subchronic Neurotoxicity Study

53057-0030; 237963; "BAS 800 H - Repeated Dose 90-Day Oral Neurotoxicity Study in Wistar Rats; Administration in the Diet'; (U. Kaspers, V. Strauss, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 63S0414/01198; 7/31/07); Ten Wistar rats rats/sex/group (unless noted otherwise) received 0, 50, 250, 1000 (males only), or 1350 ppm (females only) ((M) 0, 3.3, 16.5, 66.2 mg/kg/day, (F) 0, 3.9, 19.4, 101.0 mg/kg/day). The mean body weight gains of the 1000 ppm males and the 1350 ppm females were less than the control values (NS, $p<0.01$ or 0.05). The mean food consumption of the 1000 ppm males and the 1350 ppm females was less than that of the controls over the course of the study (NS, $p<0.01$ or 0.05). No apparent treatment-related effects were evident in the FOB or motor activity assessments. In the hematology evaluation, the mean hemoglobin concentrations, hematocrit values and corpuscular volumes of the males in the 1000 ppm group and the females in the 1350 ppm group were less than those of the control animals (NS, $p<0.01$ or 0.05). The mean corpuscular hemoglobin values of the males in the 250 and 1000 ppm groups and the females in the 1350 ppm group were less than the control values ($p<0.01$or 0.05). The mean corpuscular hemoglobin concentrations of the 250 and 1000 ppm males were less than that of the control animals ($p<0.01$ or 0.05). The mean platelet count of the 1350 ppm females was greater than that of the control group ($p<0.01$). No lesions were noted in the histopathogical examination of the nervous tissue. **No neurotoxic adverse effect was noted. Possible adverse effect:** anemia. **Reported Rat Subchronic Neurotoxicity NOEL:** (M) 1000 ppm (66.2 mg/kg/day) (based on the lack of neurotoxic effects on the 1000 ppm males), (F) 1350 ppm (101.0 mg/kg/day (based on lack of neurotoxic effects on the 1350 ppm females); **Rat Subchronic Dietary Toxicity NOEL:** (M) 50 ppm (3.3 mg/kg/day) (based on the treatment-related effects on the hematology of the 250 ppm males), (F) 250 ppm (101.0 mg/kg/day) (based on treatment-related effects on the hematology of the 1350 ppm females) **Study unacceptable**, possibly upgradeable to acceptable with the submission of concurrent positive control study data**.** (Moore, 5/23/08)

SUBCHRONIC TOXICITY STUDIES

Rat 4-Week Dietary Toxicity Study

53057-0023; 237955; "BAS 800 H - Range Finding Study in Wistar Rats, Administration in the Diet for 4 Weeks"; (U. Kaspers, V. Strauss, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 30S0414/01147; 6/6/07); Five Wistar rats/sex/group received 0, 50, 150, 450, 1350 or 4050 ppm of BAS 800 H (batch no. COD-000227; purity: 94.2%) in the diet for 4 weeks ((M) 0, 4.5, 13.4, 39.2, 116.8 and 357.2 mg/kg/day, (F) 0, 5.0, 15.0, 43.6, 130.4 and 375.6 mg/kg/day). No deaths resulted from the treatments. The males in the 1350 and 4050 ppm groups exhibited slight to moderate paleness of the skin. Only the females in the 4050 ppm group exhibited slight

paleness of the skin. The males in the 450, 1350 and 4050 ppm groups and the females in the 1350 and 4050 ppm groups had discolored urine. The mean body weight gain of the 1350 and 4050 ppm males was less than that of the control animals ($p<0.01$). There was no apparent treatment-related effect on the food consumption. The ophthalmological examination did not reveal any treatment-related effects. In the hematology evaluation, the mean white blood cell counts of both sexes in the 4050 and the males in the 1350 ppm groups were greater than the control values ($p<0.01$). The mean red blood cell counts of both sexes in the 4050 ppm group and the males in the 1350 ppm group were less than the control values ($p<0.01$). The mean hemoglobin concentrations of both sexes in the 1350 and 4050 ppm groups and the 450 ppm males were less than those of the control animals ($p<0.01$ or 0.05). The mean hematocrit values for both sexes in the 1350 and 4050 ppm groups were less than the control values ($p<0.01$ or 0.05). The mean corpuscular volume and corpuscular hemoglobin of both sexes in the 1350 and 4050 ppm groups and the males in the 450 ppm group were less than those of the control ($p<0.01$ or 0.05). The mean corpuscular hemoglobin concentrations of both sexes in the 4050 ppm group and the males in the 1350 ppm group were less than the control values ($p<0.01$or 0.05). The mean platelet count of the 4050 ppm females was greater than that of the control group ($p<0.01$). The mean reticulocyte percentages of both sexes in the 4050 ppm group were greater than those of the control group (NS). In the clinical chemistry, the serum total bilirubin values of both sexes in the 4050 ppm group and the males in the 1350 ppm group were greater than those of the control group ($p<0.01$). The total protein and albumin concentrations in the serum of both sexes in the 4050 ppm group were less than those of the control group ($p<0.01$ or 0.05). The mean globulin concentrations of the males in the 1350 and 4050 ppm group were also less than the control values ($p<0.01$ or 0.05). In the urinalysis, increased levels of urobilinogen were noted for 4 males in the 150 ppm group and all of the other males in the higher treatment groups. For the females, the urobilinogen levels were increased for 3 animals in the 450 ppm group, for 4 animals in the 1350 ppm group and for all of the animals in the 4050 ppm group. In the necropsy, the mean absolute adrenal weights of the 150 ppm males and above were less than that of the control ($p<0.01$ or 0.05). However, the mean relative adrenal weights of the 1350 and 4050 ppm groups were not significantly different from the control value and no lesions were noted in the histological examination. The mean absolute and relative spleen weights of both sexes in the 4050 ppm group and the males in the 1350 ppm group were greater than those of the control ($p<0.01$). The mean relative heart weight of the 4050 ppm males was greater than the control value ($p<0.05$). In the histological examination, erythroid hyperplasia was noted in the bone marrow of both sexes in the 4050 ppm group ((M/F): 0: 0/5 vs. 4050: 4/5). Marked extramedullary hematopoiesis was noted in the spleens of both sexes in the 4050 ppm group and in the males of the 1350 ppm group ((M) 0: 0/5 vs. 1350 and 4050: 5/5, (F) 0: 0/5 vs. 4050: 5/5). Extramedullary hematopoiesis was also evident in the livers of both sexes in the 4050 ppm group and the males of the 1350 ppm group ((M) 0: 0/5 vs. 1350 and 4050: 5/5, (F) 0: 0/5 vs. 4050: 5/5). In conjunction with the breakdown of the red blood cells, increased iron storage was evident in the liver of both sexes in the 4050 ppm group and the males of the 1350 ppm group ((M) 0: 0/5 vs. 1350 and 4050: 5/5, (F) 0: 0/5 vs. 4050: 5/5). Target organs: erythropoietic homeostasis in the bone marrow, spleen and liver. **Possible adverse effect:** anemia. **Rat 4-Week Dietary Toxicity NOEL:** (M) 50 ppm (4.5 mg/kg/day) (based on the presence of urobilinogen in the urine of the 150 ppm males), (F) 150 ppm (15.0 mg/kg/day (based on the presence of urobilinogen in the urine of the 450 ppm females); **Study supplemental** (non-guideline study). (Moore, 5/12/08)

53057-0041; 237979; "BAS 800 H - Comparative Bioavailability/Toxicity Study in Male Wistar Rats for the Hydrate and Anhydrate Crystalline Forms"; (G. Cunha, W. Mellert, K. Deckhardt, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 48C0414/01165; 11/18/05); Ten male CrlGlxBrlHan:WI rats/group received 0 or 1350 ppm of either BAS 800 H-hydrate (batch no. COD-000298, purity: 93.9%) or BAS 800 H-anhydrate (batch no. 31896/082, purity: 99%) in the diet for 4 weeks. No deaths resulted from the treatments. The mean body weight gain of both treatment groups was less than that of the control group ($p<0.05$). The mean hemoglobin values, the mean hematocrits, the mean corpuscular volumes, mean corpuscular hemoglobin values and the mean corpuscular hemoglobin concentrations of both treated groups were less than the control values ($p<0.01$). The total porphyrin levels in the feces and livers were greater in the two

treatment groups than in the control group (p<0.01). **Possible adverse effect:** anemia; **No NOEL assigned; Study supplemental.** (Moore, 8/12/08)

Rat Subchronic Dietary Toxicity Study

53057-0025; 237958; "BAS 800 H - Repeated Dose 90-Day Oral Toxicity Study in Wistar Rats - Administration in the Diet"; (U. Kaspers, V. Strauss, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 50S0414/01156; 5/2/07); Ten CrlGlxBrlHan:WI rats/sex/group (unless noted otherwise) received 0, 50, 150, 450 (males only), 1350 or 4050 ppm (females only) in the diet for 90 days ((M) 0, 3.5, 10.5, 32.5, 94.7 mg/kg/day, (F) 0, 4.3, 12.6, 110.5, 344.7 mg/kg/day). Two females in the 4050 ppm group were found dead on study day 53. The remaining females in this group were euthanized at this time for humane reasons. The mean body weight gains of the 1350 ppm males and the 4050 ppm females (up to day 49) were less than the control values (p<0.01 or 0.05). The mean food consumption of the 1350 ppm males was less than that of the controls over the course of the study. No treatment-related effect was noted in the ophthalmological examination. No apparent treatment-related effects were evident in the FOB. In the hematology evaluation, the mean white blood cell counts of both sexes in the 1350 ppm group were greater than the control values (p<0.01 or 0.05). The mean hemoglobin concentrations, hematocrit values, mean corpuscular volumes and mean corpuscular hemoglobins of both sexes in the 1350 ppm group and the 450 ppm males were less than those of the control animals (p<0.01). The mean corpuscular hemoglobin concentrations of both sexes in the 1350 ppm group were less than the control values (p<0.01or 0.05). The mean platelet count of the 1350 ppm females was greater than that of the control group (p<0.05). The mean reticulocyte percentages of both sexes in the 1350 ppm group were greater than those of the control group (NS). In the clinical chemistry, the serum total bilirubin value of the males in the 1350 ppm group was greater than the control value (p<0.01). The total protein concentrations in the serum of the males in the 150, 450 and 1350 ppm groups were less than that of the control group (p<0.01 or 0.05). The mean globulin concentrations of the males in the 450 and 1350 ppm groups were also less than the control values (p<0.01 or 0.05). No treatment-related effects were apparent in the clinical chemistry evaluation of the females. In the urinalysis, increased levels of urobilinogen were noted for 8 males in the 150 ppm group and all of the males in the higher treatment groups. Bilirubin was increased for 7 and 8 males, respectively, in the 450 and 1350 ppm groups. An increased number of transitional epithelial cells were found in the urine of 9 males in the 450 ppm group and in all 10 males of the 1350 ppm group. Granulated casts were also found in the urine of all 10 males in the 1350 ppm group. For the females, the urobilinogen levels were increased for 9 animals in the 1350 ppm group. Three of these females also had elevated levels of bilirubin.

The mean absolute and relative spleen weights of the males in the 1350 ppm group and the mean relative spleen weight of the 450 ppm males were greater than those of the control (p<0.01 or 0.05). The mean absolute and relative heart weights of the 1350 ppm males and the mean relative heart weight of the 450 ppm males were greater than the control values (p<0.01 or 0.05). The mean relative testes weights of the 450 and 1350 ppm males were greater than the control value (p<0.01 or 0.05). The mean relative liver weight of the 1350 ppm females was greater than that of the control animals (p<0.05). No lesions were noted in the heart or the testes in the histological examination. Marked extramedullary hematopoiesis was noted in the spleens of both sexes in the 1350 ppm group and in the males of the 450 ppm group ((M) 0: 1/10 vs. 450: 4/10, 1350: 10/10, (F) 0: 1/10 vs. 1350: 5/10). Extramedullary hematopoiesis was also evident in the livers of both sexes in the 1350 ppm group ((M) 0: 0/10 vs. 1350: 5/10, (F) 0: 0/5 vs. 1350: 2/10). Increased iron storage was evident in the livers of the 450 and 1350 ppm males (0: 0/10 vs. 450: 2/10, 1350: 10/10). Target organs: erythropoietic homeostasis in the spleen and liver. **Possible adverse effect:** anemia. **Rat Subchronic Dietary Toxicity NOEL:** (M) 50 ppm (3.5 mg/kg/day) (based on the increased level of urobilinogen in the urine of the 150 ppm males), (F) 150 ppm (43.6 mg/kg/day (based on treatment-related effects on the hematology and increased incidence of extramedullary hematopoiesis in the livers and spleen of the 1350 ppm females); **Study acceptable.** (Moore, 5/15/08)

Rat 28-Day Repeated Dosing Dermal Toxicity Study

53057-0029; 237962; "Repeat Dose 28-Days Dermal Toxicity Study in Wistar Rats"; (U. Kaspers, V. Strauss, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen, Germany; Report No. 33S0414/01193; 8/30/06); The skin of 10 Wistar rats/sex/group was exposed to 0 (vehicle: aqueous 0.5% carboxymethyl-cellulose), 100, 300 or 1000 mg/kg/day of BAS 800 H (batch no. COD - 000515, purity: 93.8%) for 6 hours/day, 5 days per week for 4 weeks. No treatment-related effect was noted for the mean body weight gain or food consumption. No localized dermal irritation was evident at the application site. The FOB, hematology and clinical chemistry evaluations and ophthalmology examination did not reveal any treatment-related effects. In the urinalysis, 9 of ten males and 6 of 10 females in the 1000 mg/kg group and 5 of 10 females in the 300 mg/kg group had urobilinogen scores of $\geq$ 2 ($p<0.01$ or 0.05). Six of 10 females in the 1000 mg/kg group also had a score of 3 for crystals ($p<0.05$). In the necropsy examination, no treatment-related effect on organ weights was evident. No treatment-related lesions were noted in the histopathological examination. **No adverse effect indicated. Rat Repeated Dose 28-Day Dermal Toxicity NOEL: (M)** 300 mg/kg/day (based upon increased urobilinogen levels in the urine of the 1000 mg/kg males), **(F)** 100 mg/kg/day (based upon increased urobilinogen levels in the urine of the 300 mg/kg females). **Study acceptable.** (Moore, 5/22/08)

Mouse 4-Week Dietary Toxicity Study

53057-0024; 237957; "BAS 800 H - Range Finding Study in C57BL/6NCrL Mice, Administration in the Diet for 4 Weeks"; (U. Kaspers, V. Strauss, W. Kaufmann, E. Fabian, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 31S0414/01148; 5/11/07); Five C57BL/6NCrL mice/sex/group received 0, 50, 150, 450, 1350 or 4050 ppm of BAS 800 H (batch no. COD-000227; purity: 94.2%) in the diet for 4 weeks ((M) 0, 12.8, 36.6, 112.0, 335.2 and 882.4 mg/kg/day, (F) 0, 17.9, 63.4, 153.1, 445.5 and 1620.9 mg/kg/day). One female in the150 ppm group was found dead on day 28. The mean body weight gains of the 450, 1350 and 4050 ppm males were less than that of the control animals ($p<0.01$or 0.05). There was no apparent treatment-related effect on the food consumption. In the hematology evaluation, the mean red blood cell counts of the males in the 1350 and 4050 ppm groups were less than the control value ($p<0.01$). The mean hemoglobin concentrations of both sexes in the 1350 and 4050 ppm groups and the 150 and 450 ppm males were less than those of the control animals ($p<0.01$ or 0.05). The mean hematocrit values for both sexes in the 4050 ppm groups and the males in the 150, 450 and 1350 ppm groups were less than the control values ($p<0.01$ or 0.05). The mean corpuscular volume of both sexes in the 4050 ppm groups and the males in the 450 and 1350 ppm groups were less than those of the control ($p<0.01$). The mean corpuscular hemoglobin of both sexes in the 1350 and 4050 ppm groups and the males in the 450 ppm group were less than those of the control ($p<0.01$ or 0.05). The mean corpuscular hemoglobin concentrations of the females in the 1350 and 4050 ppm groups were less than the control value ($p<0.01$). The mean reticulocyte percentages of both sexes in the 4050 ppm group were greater than those of the control group (NS). In the clinical chemistry, the mean serum alanine and aspartate aminotransferase activities of the males in the 150 ppm group and above were greater than the control value ($p<0.01$ or 0.05). The alkaline phosphatase activity in the serum of the 4050 ppm males was greater than the control value ($p<0.01$). The serum urea and total bilirubin concentrations of the 150 ppm males and above were greater than the control value ($p<0.01$or 0.05). The mean serum alanine aminotransferase activities of the females in the 1350 and 4050 ppm groups were greater than the control value ($p<0.01$). In the necropsy, the mean absolute and relative liver weights of both sexes in the 1350 and 4050 ppm groups and the males in the 150 and 450 ppm groups were greater than the control values ($p<0.01$). The mean absolute and relative spleen weights of the males in the 4050 ppm group were greater than the control values ($p<0.01$). The mean relative thymus weight of the 4050 ppm males was greater than the control value ($p<0.01$). In the histological examination, marked extramedullary hematopoiesis (grades 3 or 4) was noted in the spleens of both sexes in the 4050 ppm group and in the males of the 1350 ppm group ((M) 0: 0/5 vs. 1350: 4/5, 4050: 5/5, (F) 0: 0/5 vs. 4050: 5/5). Extramedullary hematopoiesis was evident in the livers of the males in the 450, 1350 and 4050 ppm groups (0: 0/5 vs. 450: 5/5, 1350: 4/5 and 4050: 5/5). Centrilobular fatty change (grade 2 or greater) was noted in the livers of both sexes in the 450, 1350, and 4050 ppm groups and the males in the 150 ppm group ((M) 0: 0/5 vs. 150, 450, 1350 and 4050: 5/5, (F)

0: 0/5 vs. 450, 1350 and 4050: 5/5). Target organs: erythropoietic functions in the liver and spleen. **Possible adverse effect:** anemia. **Rat 4-Week Dietary Toxicity NOEL:** (M) 50 ppm (12.8 mg/kg/day) (based on treatment-related effects on the hematology, clinical chemistry and liver of the 150 ppm males), (F) 150 ppm (63.4 mg/kg/day (based on centrilobular fatty change in the livers of the 450 ppm females); **Study supplemental** (non-guideline study). (Moore, 5/14/08)

Mouse Subchronic Dietary Toxicity Study

53057-0026; 237959; "BAS 800 H - Repeated Dose 90-Day Oral Toxicity Study in C57BL/6NCrL Mice, Administration in the Diet"; (U. Kaspers, V. Strauss, W. Kaufmann, E. Fabian, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 51S0414/01161; 6/14/07); Ten C57BL/6NCrL mice/sex/group (unless noted otherwise) received 0, 15 (males only) 50, 150, 450, or 1350 ppm (females only) ((M) 0, 3.6, 12.5, 36.7, 109.1 mg/kg/day, (F) 0, 17.6, 51.8, 156.7, 471.2 mg/kg/day). No deaths occurred during the study. The mean body weight gains of the 150 and 450 ppm males were less than the control values (NS). There was no apparent treatment-related effect on food consumption. In the hematology evaluation, the mean hemoglobin concentrations, hematocrit values, mean corpuscular volumes, mean corpuscular hemoglobin values and mean corpuscular hemoglobin concentrations of the 150 and 450 ppm males and the 1350 ppm females were less than the control values ($p<0.01$). Although some of these hematological parameters for the other treatment groups were statistically different from the control values, the effects were not deemed to be physiologically significant. In the clinical chemistry evaluation, the mean serum alanine aminotransferase activities of the males in the 150 and 450 ppm groups were greater than the control value ($p<0.01$). The serum aspartate aminotransferase and the alkaline phosphatase activities of the 450 ppm males were elevated as well ($p<0.01$ or 0.05). The serum urea and total bilirubin concentrations of the 450 ppm males were greater than the control values ($p<0.01$ or 0.05). For the 450 and 1350 ppm females, the mean serum albumin levels were less than the control value ($p<0.01$ or 0.05). The mean absolute and relative liver weights of the 150 and 450 ppm males and the 1350 ppm females were greater than the control values ($p<0.01$). The mean relative liver weight of the 450 ppm females was also greater than that of the control ($p<0.05$). The mean relative kidney weights of the 150 and 450 ppm males were greater than that of the control group ($p<0.01$ or 0.05). In contrast, the mean relative kidney weights of the 450 and 1350 ppm females were less than the control value ($p<0.01$). The increased weight noted for the kidneys in the males correlated with the increased level of urea in the serum. In the histopathological examination, increased lymphoid infiltration was noted in the livers of the 150 and 450 ppm males and the 450 and 1350 ppm females ((M) 0: 0/10 vs. 150: 7/10, 450: 10/10, (F) 0: 0/10 vs. 450: 3/10, 1350: 10/10). Marked diffuse fatty change (grade $\geq$3) was noted in the livers of the 150 and 450 ppm males (0: 0/10 vs. 150: 8/10, 450: 10/10). Marked central fatty change (grade $\geq$3) was evident in the livers of the 450 and 1350 ppm females (0: 0/10 vs. 450: 8/10, 1350: 10/10). Target organ: liver. **Possible adverse effect:** fatty change in the liver. **Mouse Subchronic Dietary Toxicity NOEL: (M)** 50 ppm (12.5 mg/kg/day) (based on the treatment-related effects on the hematology and clinical chemistry parameters and the increased absolute and relative liver weights and hepatic lesions noted for the 150 ppm males), **(F)** 150 ppm (51.8 mg/kg/day (based on treatment-related effects on the relative liver weights and hepatic lesions noted for the 450 ppm females); **Study supplemental** (non-guideline study). (Moore, 5/19/08)

Dog 4-Week Oral Toxicity Study

53057-0027; 237960; "BAS 800 H - Subacute Oral Toxicity Study in Beagle Dogs, Administration Via Gelatin Capsules for 4 Weeks (Including Amendments 1, 2, & 3)"; (U. Kaspers, K. Deckardt, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 40D0414/01164; 11/14/05 (amend. no. 1, 11/24/05, amend. no. 2, 11/20/06, amend. no. 3, 3/29/07); Four beagle dogs/sex/group received 0, 30, 100, or 300 mg/kg/day of BAS 800 H (batch no. COD-000515, purity: 93.8%) orally, in capsules, for 4 weeks. No deaths occurred during the study. No treatment-related effects on mean body weight gain or food consumption were evident. In the clinical observations, the feces of both sexes in the 100 and 300 mg/kg groups was discolored dark brown and/or reddish brown. The onset was within 4 days of study initiation. In the

hematological evaluation, the mean hemoglobin concentration and hematocrit percentages for both sexes in the 300 mg/kg group were less than the control values (NS, p<0.05). The mean corpuscular volume and corpuscular hemoglobin values of both sexes in the 100 and 300 mg/kg groups were less than those of the controls (p<0.05). The mean platelet number of both sexes in the 300 mg/kg group and the males in the 100 mg/kg group were greater than the control values (p<0.05). The activated partial thromboplastin time (PTT) of both sexes in the 100 and 300 mg/kg groups was less than the control group (NS, p<0.05). In the clinical chemistry evaluation, the serum alkaline phosphatase activities of both sexes in the 300 mg/kg group and the females in the 100 mg/kg group were elevated above the control values (p<0.05). The serum albumin concentrations of both sexes in the 300 mg/kg group were less than those of the controls (NS, p<0.05). The porphyrin concentrations in the plasma and feces of both sexes in the 30 mg/kg group and above were greater than the control values (p<0.05). The concentration of porphyrins in the urine were also elevated for both sexes in the 100 and 300 mg/kg groups (p<0.05). The mean absolute and relative liver weights of both sexes in the 300 mg/kg group and the males in the 100 mg/kg group were greater than the control values (NS). The mean absolute and relative spleen weights of both sexes in the 300 mg/kg group and the females in the 100 mg/kg group were greater than the control values (NS). In the histopathological examination, iron storage in the liver was evident for one male in the 100 mg/kg group and for 2 males and 3 females in the 300 mg/kg group. In the spleen, a greater degree of iron storage (≥ grade 2) was noted for the males in the 100 and 300 mg/kg groups (0: 0/4 vs. 100: 4/4, 300: 2/4). A similar effect was not apparent for the females. Extramedullary hematopoiesis was noted in the spleens of both sexes in the 300 mg/kg group and the males in the 100 mg/kg group ((M) 0: 0/4 vs. 100: 1/4, 300: 2/4, (F) 0: 0/4 vs. 300: 4/4). Hyperplasia in the bone marrow of both sexes in the 300 mg/kg group and the females in the 100 mg/kg was reported ((M) 0: 0/4 vs. 300: 4/4, (F) 0: 0/4 vs. 100: 1/4, 300: 4/4). **Possible adverse effect:** perturbation of erythropoiesis in the liver, spleen and bone marrow; **Dog 4-Week Oral Toxicity NOEL:** (M/F) < 30 mg/kg/day (based upon the increased levels of porphyrins recovered in the plasma and feces of both sexes in the 30 mg/kg group); **Study supplemental** (non-guideline). (Moore, 5/21/08)

Dog Subchronic Oral Toxicity Study

53057-0028; **237961**; "BAS 800 H - Repeated 90-Day Oral Toxicity Study in Beagle Dogs, Administration via Gelatin Capsules"; (U. Kaspers, K. Deckardt, S. Burkhardt, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 41D0414/01182; 4/5/06); Five beagle dogs/sex/group received 0, 10, 50, or 150 mg/kg/day of BAS 800 H (batch no. COD-000606, purity: 93.8%) orally, in capsules, for 13 weeks. No deaths occurred during the study. The mean body weight gains of both sexes in the 150 mg/kg treatment group were less than the control values over the course of the study (NS). The food consumption of the males did not demonstrate a treatment-related effect. The females in the 150 mg/kg group consumed less food than did the control group. In the clinical observations, dark red-brown to dark brown feces were noted for both sexes in the 150 mg/kg group (onset by day 6). The ophthalmological examination did not reveal any treatment-related effects. In the hematological evaluation, the mean hemoglobin concentrations of the males in the 150 mg/kg group were less than the control values throughout the study (p<0.05 and 0.01). The mean hematocrit percentage of the 150 mg/kg males was less than that of control at the termination of the study (p<0.01). The mean corpuscular volume and corpuscular hemoglobin values of both sexes in the 50 and 150 mg/kg groups were less than those of the controls at 6 weeks and/or termination of the study (p<0.01 or 0.05). The mean corpuscular hemoglobin concentration values for both sexes of the 150 mg/kg group were less than the control values throughout the study (p<0.01 or 0.05). The mean platelet number of both sexes in the 150 mg/kg group were greater than the control values throughout the study (NS, p<0.05). In the clinical chemistry evaluation, the serum alkaline phosphatase activities of both sexes in the 150 mg/kg group and the females in the 50 mg/kg group were elevated above the control values throughout the study (p<0.01 or 0.05). The serum albumin concentrations of both sexes in the 150 mg/kg group and the males in the 50 mg/kg group were less than those of the controls throughout the study (p<0.01). The mean absolute and relative liver weights of the males in the 150 mg/kg group were greater than the control values (NS, p<0.05). The mean absolute and relative spleen weights of the females in the 150 mg/kg group were greater than the control

values (NS). In the histopathological examination, increased iron storage in the liver was evident for one male and one female in the 50 mg/kg group and for 3 males and 4 females in the 150 mg/kg group. In the spleen, a greater degree of iron storage (≥ grade 2) was noted for the males in the 10, 50 and 150 mg/kg groups (0: 0/5 vs. 10: 1/5, 50: 2/5, 150: 4/5) and for the females in the 50 and 150 mg/kg groups (0: 0/5 vs. 50: 3/5, 150: 5/5). Extramedullary hematopoiesis was noted in the spleens of the females in the 50 and 150 mg/kg groups (0: 0/5 vs. 50: 1/5, 150: 2/5). Hyperplasia in the sternal bone marrow of both sexes in the 150 mg/kg group was reported ((M and F) 0: 0/5 vs. 150: 2/5, note: femoral bone marrow from the 150 mg/kg females also exhibited hyperplasia). **Possible adverse effect:** perturbation of erythropoiesis in the liver, spleen and bone marrow; **Dog Subchronic Oral Toxicity NOEL: (M)** < 10 mg/kg/day (based upon the increased level of iron storage in the spleen of the10 mg/kg males), **(F)**: 10 mg/kg/day (based upon treatment-related effects noted in the hematology evaluation and increased iron storage in the liver and spleen and the presence of extramedullary hematopoiesis in the spleen of the 50 mg/kg females). **Study acceptable**. (Moore, 5/22/08)

METABOLISM STUDIES

Metabolism, Rat

** 53057-0038; 237975; "^{14}C-BAS 800 H: Study on the Biokinetics in Rats"; (E. Fabian, R. Landsiedel; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 02B0627/046008; 9/24/07); Wistar rats of both sexes were dosed orally by gavage with (Phenyl-^{14}C)-BAS 800 H (batch no. 825-1085, radiochemical purity: >95.0%, chemical purity: 100%, specific activity: 5.54 MBq/mg or batch no. 825-1201, radiochemical purity: >95.0%, chemical purity: 92.5%, specific activity: 5.19 MBq/mg) in 1) Blood/Plasma, 2) Balance/Excretion, 3) Tissue Distribution and 4) Bile Excretion studies. In addition, (uracil-5-^{13}C), (benzymide-carbonyl-^{13}C)-BAS 800 H, (batch no. 828-1085, chemical purity: 99%) was used in the Bile/Excretion Study. Unlabeled BAS 800 H (batch no. COD-000298, chemical purity: 93.9%) was used to adjust the concentration of the test material in dosing preparations for the required specific activity or in the multiple dose regimen for the Balance/Excretion study. In the Blood/Plasma study, 4 animals/sex/group received a single dose of 4, 20 or 100 mg/kg of the test material and blood samples were recovered up to 7 days post-dose. In the Balance/Excretion study, 4 animals/sex/group received either a single dose of 5 or 100 mg/kg of the radiolabeled test material or 100 mg/kg/day of the unlabeled test material for 14 days followed by a single dose of 100 mg/kg of the radiolabeled test material. Urine and feces were collected up to 7 days post-dose (final dose). In the Tissue Distribution study, 12 animals/sex/group were dosed with 5 or 100 mg/kg of the test material. Specified tissues were analyzed for radioactivity at the times for which the plasma concentration was maximal, 1/2 the maximal level, 1/4 the maximal level and 1/8 the maximal level. For the 100 mg/kg treatment group, 3 animals/sex/time point were euthanized at 1, 7, 20 and 34 hours post-dose. For the 5 mg/kg group, 3 males/time point were euthanized at 1, 24, 48 and 72 hours post-dose and 3 females/time point were euthanized at 1, 4, 20 and 24 hours post-dose. In the Biliary Excretion study, 4 bile-duct cannulated animals/sex/group were dosed with 5 or 100 mg/kg of the test material and bile samples were collected at 3 hour intervals up to 48 hours post-dose. In the Balance/Excretion Study, there was a sex-specific difference in the excretion profile. The urine was the primary route of excretion for the females in which 84 to 96% of the administered dose was recovered over the 5 to 100 mg/kg dosing range. In contrast, 26% of the administered dose was recovered in the urine of the 5 mg/kg males, increasing to 53 and 62% for the 100 mg/kg (single dose) males and the 100 mg/kg/day (multiple dose) males, respectively. Radiolabel recovered in the feces ranged from 10% of the administered dose for the 100 mg/kg females (single dose) to 81% for the 5 mg/kg males. The percentage of the administered radiolabel which was excreted within 48 hours of dosing ranged from 82.5% (5 mg/kg males) to 101.3% (5 mg/kg females). In the Blood/Plasma study, maximal plasma levels for radiolabel were achieved within 1 hour of dosing. The maximal concentrations in the plasma were 23 to 24 ug equivalents/gram of plasma for the 4 mg/kg group, 85 to 98 ug equivalents/ gram of plasma for the 20 mg/kg group and 258 to 286 ug equivalents/gram of plasma for the 100 mg/kg group. The initial half lives in the plasma ranged from 4.9 hours (100 mg/kg females) to 20.9 hours (4 mg/kg males). The terminal half lives ranged from 20.9 hours (4 mg/kg males) to 59.2 hours (100 mg/kg females). The area under the curve was proportionately greater for the males than the females (4 mg/kg:

741 to 247 ug equivalents * hr/gram, 20 mg/kg: 2131 to 754 ug equivalents * hr/gram and 100 mg/kg: 4502 to 3057 ug equivalents * hr/gram). In the Tissue Distribution study, none of the radiolabel was sequestered in any particular tissue. In the Biliary Excretion study, a sex-specific effect was noted. In the males, 52 and 68% of the administered radiolabel was recovered in the bile of the 5 and 100 mg/kg males, respectively, up to 48 hours post-dose. In contrast, 18 and 35% of the administered radiolabel was recovered in the bile of the 5 and 100 mg/kg females, respectively, over the same time period. Absorption of the radiolabel was virtually complete for the females in the 5 mg/kg group and both sexes in the 100 mg/kg group (the percentage of the administered dose recovered in the urine and bile was greater than 100%). For the males in the 5 mg/kg group, 78% of the dose was recovered in the urine and bile. **Study acceptable.** (Moore, 7/22/08)

** 53057-0039; 237976; "The Metabolism of ^{14}C-BAS 800 H (Reg No. 4054449) in Rats'; (F. Grosshans; BASF Aktiengesellschaft, BASF Agricultural Center Limburgerhof, Crop Protection Division, Ecology and Environmental Analytics, 672114 Limburgerhof, Germany; Report No. 132617; 10/17/07); Wistar rats of both sexes were dosed orally by gavage with (phenyl-^{14}C)-BAS 800 H (batch no. 825-1085, radiochemical purity: >95.0%, chemical purity: 100%, specific activity: 5.54 MBq/mg or batch no. 825-1201, radiochemical purity: >95.0%, chemical purity: 92.5%, specific activity: 5.19 MBq/mg or (uracil-4-^{14}C)-BAS 800 H, batch no. 829-1017, radiochemical purity: 99.5%, chemical purity: 99.2%, specific activity: 4.26 MBq/mg) in 1) Balance/Excretion and 2) Tissue Distribution studies. Unlabeled BAS 800 H (batch no. L67-140, purity: 99.9% or batch no. COD-000298, chemical purity: 93.9%) was used to adjust the concentration of the test material in dosing preparations for the required specific activity. In the Balance/Excretion study, animals were treated with 100 mg/kg of the test material and urine and fecal samples were collected up to 4 days post-dose (except for the females treated with the uracil-labeled test material in which samples were collected for up to 7 days post-dose). For the Tissue Distribution study, animals were treated with either 5 or 100 mg/kg of the test material and liver, kidney, fat and plasma samples were recovered 1 hour post-dose. Samples were analyzed for total radioactivity and aliquots of each sample were fractionated by HPLC and the chemical structure of the isolated moieties was elucidated by mass spectroscopy. In addition, urinary, fecal and biliary samples recovered from the Balance/Excretion and Biliary Excretion studies, cited in Report No. 02B0627/046008, (vol. no. 53057-0038, rec. no. 237975), were subjected as well to HPLC and mass spectroscopy. As observed in the previous study, there was a sex-specific difference in the balance/excretion study in this report. The primary route of excretion for the males was the feces (47 to 50% vs. 28 to 29% in the urine). For the females, 60 to 62% of the administered dose was recovered in the urine and cage wash in contrast to 14 to 15% in the feces. The position of the labeling on the phenyl or uracil moiety did not affect the excretion profile. In the tissue distribution study, 24 to 36% of the administered dose was recovered in the liver, kidney, fat or plasma of the 5 mg/kg animals at 1 hour post-dose. For the 100 mg/kg animals, the percentage of the administered dose which was recovered in these tissues at 1-hour post-dose declined to 8 to 13%. In the profiles of the metabolites, a sex-specific difference was noted. In the urine of the males (collected in the previous study), the primary radiolabeled moieties were the parent compound, M800H01, M800H03 and M800H05. Demethylation and depropylation of the sulfonamide-N-methyl-N-isopropyl group occurred in a step-wise process. These moieties represented 91 to 97% of the total recovered radioactivity. An additional metabolite M800H07 was produced by cleavage of the uracil ring with the loss of 3 carbon atoms. For the animals treated with the uracil-labeled test material, this metabolite was not detectable. For the females, the parent compound represented 93 to 95% of the radiolabel which was recovered in the urine. In the excretion/balance data produced in this study, these results were confirmed. In the feces of both sexes, the parent compound, M800H01, M800H03 and M800H05 represented 78 to 82% of the recovered radiolabel. For the females, these moieties comprised 70 to 77% of the recovered radiolabel. Additional metabolites, M800H02, M800H04, M800H06 and M800H08, were also identified in the feces of both sexes. M800H02 resulted from a demethylation of the uracil nitrogen. Cleavage of the uracil ring resulted in the M800H04 metabolite. Reduction of the uracil ring was noted for M800H06 and the M800H08. The uracil nitrogen of the latter metabolite had also been demethylated. These metabolites represented 9 to 11% of the recovered radiolabel in the feces of both sexes. In the biliary samples, a greater

variety of metabolites were recovered. The parent compound and M800H01 represented 29 to 43% of the recovered radiolabel for both sexes at both treatment levels. M800H07 comprised another 22 to 28% of the recovered label. For the males, M800H18, a metabolite in which the uracil-ring was cleaved to form a urea in addition to being demethylated at the sulfonyl-N-methyl-N-isopropyl site, represented 24 and 14% of the recovered label in the 5 and 100 mg/kg treatment groups, respectively. For the females, this metabolite comprise only 3 to 4% of the recovered label. Additional metabolites, M800H02, M800H17 and M800H20, were also recovered in the bile of both sexes. M800H02 had been demethylated at the uracil-nitrogen site. For both M800H17 and M800H20, the uracil ring had been cleaved to form a urea with the 3-carbon fragment still attached. In addition, M800H20 had been demethylated at the sulfonyl-N-methyl-N-isopropyl site. In the tissues, the unmetabolized parent compound represented the predominant radiolabeled moiety at 1 hour post-dose. In the liver, the parent compound represented 75 to 91% of the recovered label. In the kidneys, 67 to 82% of the radiolabel was the parent compound. M800H04 comprised 6 to 22% of the recovered radiolabel in the liver and 18 to 33% in the kidneys. Metabolism of the parent compound was a stepwise process in which the N-methyl-N-isopropylsulfonamide was degraded to the unsubstituted sulfonamide and the uracil ring was cleaved with the loss of a 3-carbon fragment to form a N-methylurea attached to the phenyl ring. **Study acceptable.** (Moore, 7/28/08)

MECHANISTIC STUDIES

Rat 8-Week Dietary Toxicity and Mechanistic Studies

53057-0040; 237977; "Mechanistic Study in Wistar Rats - Total Porphyrin Analysis, Administration in the Diet for at Least 4 Weeks"; (G. Cunha, W. Mellert, V. Strauss, W. Kaufmann, L. Ma-Hock, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 30C0414/01150; 8/29/06); Ten CrlGlxBrlHan:WI rats/sex/group received 0, 10, 50 or 1000 ppm of BAS 800 H (batch no. COD-000227; purity: 94.2%) in the diet for 8 weeks ((M) 0, 0.8, 4.1, 80.6 mg/kg/day, (F) 0, 0.9, 4.6, 89.5). Five animals/sex/group were euthanized after 8 weeks of treatment. The remaining 5 animals/sex/group were maintained for an additional 2-week recovery period. Total porphyrin levels were measured at 2, 4 and 8 weeks in the plasma. Coproporphyrin III was measured in the urine and feces after 1, 2, and 4 weeks of treatment. Protoporphyrin IX and mesoporphyrin were measured in the feces after 1, 2, and 4 weeks of treatment. Total porphyrin levels were measured in the urine and feces after 2, 4, and 8 weeks of treatment; in addition, the total porphyrin level in the feces was measured after the 2-week recovery period. The δ-aminolevulinic acid and porphobilinogen levels were measured in the urine after 2 and 4 weeks of treatment. No deaths occurred during the study. The mean body weight gain of the 1000 ppm males was less than that of the control during the 1st week of treatment ($p<0.05$). The mean food consumption of the 1000 ppm males was less than that to the control animals throughout the study. In the hematology evaluation, the mean hemoglobin levels and hematocrit of both sexes in the 1000 ppm group were less than the control values after 8 weeks of treatment ($p<0.01$). No effect was evident after the 2-week recovery period. The mean corpuscular volume and mean corpuscular hemoglobin of both sexes in the 1000 ppm group were less the control values after 8 weeks of treatment ($p<0.01$). The effect was still evident for the females in this group after the 2-week recovery ($p<0.05$ or 0.01). The mean corpuscular hemoglobin concentration of the 1000 ppm males was less than that of the control males after both 8 weeks of treatment and the 2-week recovery ($p<0.05$). The total porphyrin levels in the plasma, urine and feces of both sexes in the 1000 ppm group were elevated throughout the treatment period ($p<0.01$). For both sexes in the 50 ppm group, the total porphyrin levels in the urine and feces were elevated during the treatment period as well ($p<0.01$, 0.05 or NS). The total porphyrin level in the plasma of the 50 ppm males was also elevated at 3 and 5 weeks of the treatment ($p<0.01$ or 0.05). The total porphyrin level in the feces of the 10 ppm males was greater than that of the control males during the treatment period ($p<0.01$, 0.05 or NS). The total porphyrin levels in the liver of both sexes in the 10 ppm group and above were greater than the control values after 9 weeks of treatment ($p<0.01$, 0.05 or NS). The δ-aminolevulinic acid and porphobilinogen levels in the urine of the 1000 ppm males were greater than the control values after 2 and 4 weeks of treatment ($p<0.01$ or 0.05). The porphobilinogen level of the 1000 ppm females was greater than that of the control females only

after 2 weeks of treatment ($p<0.01$). **Possible adverse effect:** anemia; **Rat Subchronic Dietary Toxicity NOEL:** (M/F) < 10 ppm ((M) <0.8 mg/kg/day, (F) <0.9 mg/kg/day) (based on the increased levels of total porphyrin in the livers of both sexes in the 10 ppm group and the feces of the males in the 10 ppm group); **Study Supplemental.** (Moore, 8/8/08)

53057-0040; 237978; " BAS 800 H - Supplementary Mechanistic Study in Wistar Rats - Total Porphyrin Analysis, Administration in the Diet over 8 Weeks"; (G. Cunha, W. Mellert, K. Deckhardt, W. Kaufmann, B. van Ravenzwaay; Experimental Toxicology and Ecology, BASF Aktiengesellschaft, 67056 Ludwigshafen/Rhein, Germany; Report No. 48C0414/01165; 10/10/05); Ten CrlGlxBrlHan:WI rats/sex/group received 0, 1, 5 or 25 ppm of BAS 800 H (batch no. COD-000227; purity: 94.2%) in the diet for 8 weeks ((M) 0, 0.1, 0.4, 2.0 mg/kg/day, (F) 0, 0.1, 0.5, 2.3). Hematology parameters and total porphyrin levels in the feces were measured at 1, 2, 4 and 8 weeks. No deaths occurred during the study. No treatment-related effect was evident on the mean body weights or food consumption. The hematology parameters were not affected by the treatment at any time during the 8-week study. For both sexes in the 25 ppm group, the total porphyrin levels in the feces were elevated during the treatment period ($p<0.01$). The total porphyrin level in the feces of the 5 ppm males was greater than that of the control males during the treatment period ($p<0.01$, 0.05). **No adverse effect noted. Rat Subchronic Dietary Toxicity NOEL:** (M) 1 ppm ((M) 0.1 mg/kg/day) (based upon the increased levels of total porphyrin in the feces of the 5 ppm males), (F) 5 ppm (0.5 mg/kg/day) (based on the increased levels of total porphyrin in the feces of the females in the 25 ppm group); **Study Supplemental.** (Moore, 8/11/08)